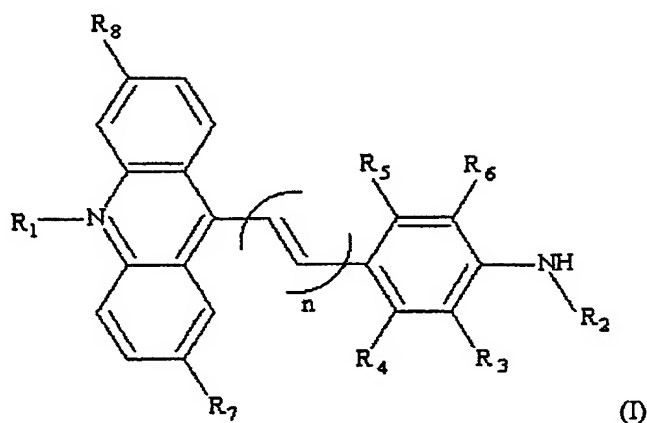


CLAIMS

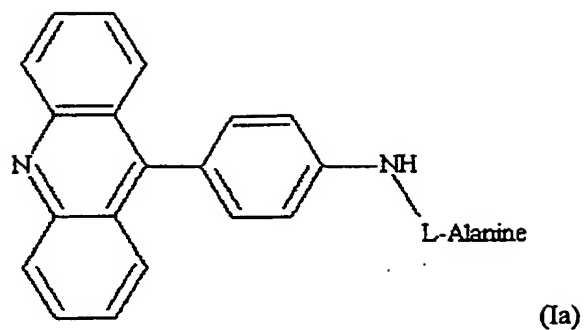
1. A chromogenic enzyme substrate for detecting aminopeptidase activity in microorganisms or for determining whether at least one bacterium belongs to the Gram-positive group or to the Gram-negative group according to the color thereof, characterized in that it has formula (I) below:



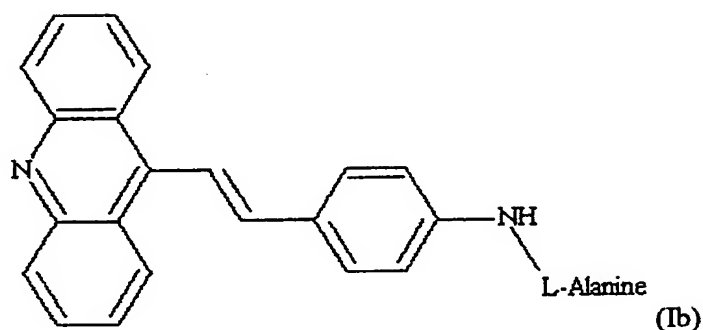
in which:

- R₁ is nothing or an alkyl, allyl or aryl group,
- R₂ consists of at least one amino acid, preferably alanine,
- R₃, R₄, R₅ and R₆ consist, independently of one another, of H- or -O-alkyl, preferably -O-CH₃,
- R₇ consists of H, O-CH₃, alkyl or halogen,
- R₈ consists of H or Cl, and
- n is an integer corresponding to 0 or 1.

2. The substrate as claimed in claim 1, characterized in that it has formula (Ia) below:



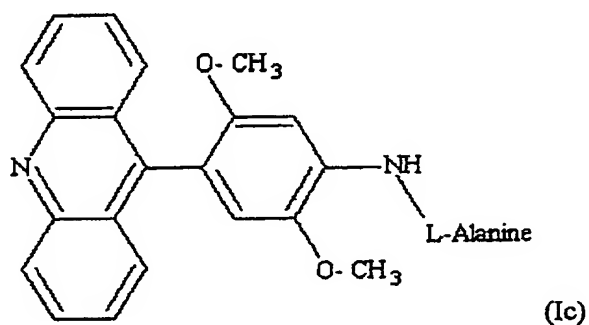
or in that it has formula (Ib) below:



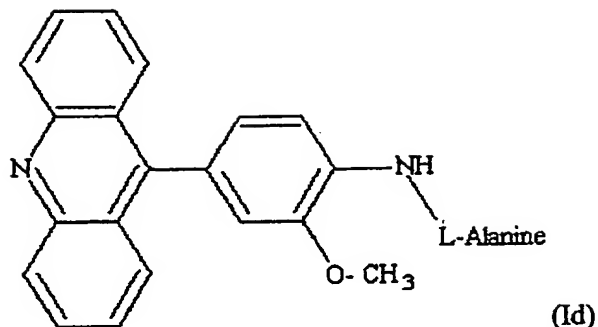
5

3. The substrate as claimed in claim 1, characterized
in that R₁ is a methyl or allyl group.

10 4. The substrate as claimed in claim 1, characterized
in that it has formula (Ic) below:



15 or in that it has formula (Id) below:



(Id)

5. The substrate as claimed in any one of claims 1 to
5 4, characterized in that R₂ or the L-alanine is coupled
to a blocking agent.

6. A culture medium using at least one chromogenic
enzyme substrate as claimed in any one of claims 1 to
10 5, alone or in combination with at least one other
enzyme substrate specific for an enzyme activity that
is other than that detected by the substrate according
to the invention.

15 7. The medium as claimed in claim 6, characterized in
that it consists of a gelled medium.

8. The use of the chromogenic enzyme substrates as
defined in any one of claims 1 to 5, or of a culture
20 medium as claimed in either one of claims 6 and 7, for
detecting at least one aminopeptidase activity in
microorganisms.

9. The use of the chromogenic enzyme substrates as
25 defined in any one of claims 1 to 5, or of a culture
medium as claimed in either one of claims 6 and 7, for
separating bacteria with Gram-positive coloration from
bacteria with Gram-negative coloration.

30 10. A method for detecting at least one aminopeptidase
activity in microorganisms, characterized in that it
consists in:

- providing a culture medium as claimed in either one of claims 6 and 7,
- seeding the medium with a biological sample to be tested,
- 5 • leaving it to incubate, and
- visualizing the presence of at least one aminopeptidase activity, alone or in combination with at least one other enzyme activity different from an aminopeptidase activity.

10

11. A method for differentiating bacteria in terms of whether they belong to microorganisms of the Gram-positive type or to microorganisms of the Gram-negative type, characterized in that it consists in:

- 15 • providing a culture medium as claimed in either one of claims 6 and 7,
- seeding the medium with a biological sample to be tested,
- leaving it to incubate, and
- 20 • visualizing the presence of at least one color synonymous with the presence of a microorganism or microorganisms of the Gram-negative type.

12. The method as claimed in either one of claims 10
25 and 11, characterized in that, when the nitrogen in the 10-position of the acridine group is not quaternized, the presence of at least one aminopeptidase activity is visualized by adding acid, preferably hydrochloric acid, acetic acid or citric acid, to the culture.